

# Quarterly Update

National Bioenergy Center Sugar Platform Integration Project



Biomass Program—Sustainable Fuels, Chemicals, Materials, and Power

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The Sugar Platform Integration Project focuses on integrating the processing steps involved in enzyme-based lignocellulose conversion technology. This project supports the U.S. Department of Energy's efforts to foster development, demonstration, and deployment of "sugar platform" biorefineries that produce inexpensive commodity sugars and fuel ethanol, as well as a variety of other fuel and chemical products, from abundant renewable lignocellulosic biomass.

The National Renewable Energy Laboratory manages this project for DOE's Office of the Biomass Program. Information on the Biomass Program is available at [Biomass Program](#)

To discuss information in this update or for further information on the Sugar Platform Integration Project, contact Dan Schell at NREL, phone (303) 384-6869, email [dan\\_schell@nrel.gov](mailto:dan_schell@nrel.gov)

## Process Integration Review Meeting Held September 15<sup>th</sup> in Washington, D.C.

Recent accomplishments and future work plans for the Process Integration Project were presented at a public meeting to a seven-member review panel, which included representatives from industry (3), academia (1), DOE headquarters (1), DOE GO (1), and NREL (1). Presentations and other materials are available for viewing at the following web site ([http://devafdc.nrel.gov/biogeneral/process\\_integration/](http://devafdc.nrel.gov/biogeneral/process_integration/)).

## 27<sup>th</sup> Symposium on Biotechnology Fuels and Chemicals Call for Papers.

The premier conference series on biotechnology for fuels and chemicals continues with the 27<sup>th</sup> Symposium this coming May in Denver, Colorado. The deadline for submission of abstracts for papers and posters for the symposium is November 30, 2004. Session topics are listed below. Please visit the web site for more information, and start preparing your abstract for submissions now.

- Session 1A Feedstock Supply and Logistics
- Session 1B Enzyme Catalysis and Engineering
- Session 2 Today's Biorefineries
- Session 3A Plant Biotechnology and Feedstock Genomics
- Session 3B Biomass Pretreatment and Hydrolysis
- Session 4 Industrial Biobased Products
- Session 5 Microbial Catalysis and Metabolic Engineering
- Session 6 Bioprocess R&D
- Special Topic A International Energy Agency Task #39-Liquid Biofuels
- Special Topic B Bioenergy Life-Cycle Analysis/Economics of Sustainability

The Symposium web site can be found at the link provided below.  
[http://www.eere.energy.gov/biomass/biotech\\_symposium/](http://www.eere.energy.gov/biomass/biotech_symposium/)

## AIChE Envisioning Biorefineries Topical Conference, Austin, Texas, November 7-12, 2004.

This topical conference, which is being held in conjunction with the AIChE annual national meeting, offers a variety of technical sessions covering advances in all areas of the renewable feedstocks processing ("biorefining") industry, including: chemical and biological processing; biorefinery stream separations; pretreatment and reactor engineering for biomass feedstocks; and life-cycle and techno-economic analyses. Recent developments in agricultural biotechnology and green bioprocessing will be described, as will progress in the manufacturing of renewable feedstock-based alternative fuels, polymers and biomaterials. See the AIChE conference web site for more details: [Envisioning Biorefineries](#)



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## R&D Progress

**New Method for Biomass Sugar Analysis.** A more accurate, higher resolution method for measuring biomass sugars using High Performance Liquid Chromatography (HPLC) has been developed under a subcontract with Professor Foster Agblevor at Virginia Polytechnic Institute and State University. Application of this method by Professor Agblevor provides the first quantifiable evidence that some of the sucrose present in the water extractives portion of corn stover can survive dilute acid pretreatment. This is an important finding, since previously it was assumed that during pretreatment all of the sucrose contained in corn stover would degrade into HMF or other degradation products. A key implication of (some) sucrose or its hydrolysis products, glucose and fructose, remaining after pretreatment is that modestly higher levels of ethanol may be able to be produced from biomass than previously assumed. The development of the new HPLC method for biomass sugar measurement made this possible. The new method improves chromatographic resolution and thereby enables more accurate quantification of a larger number of biomass-derived sugars. In addition to the five biomass sugars normally monitored in biomass hydrolysates (i.e., glucose, xylose, galactose, mannose and arabinose), the new method is able to resolve and quantify sucrose and fructose (derived from sucrose). In contrast, the methods now in routine use within the Biomass Program are not able to quantify these sugars. This was a significant limitation because corn stover (and likely other herbaceous lignocellulosic feedstocks) contains appreciable levels of sucrose. The sucrose content of corn stover is variable but significant, ranging from 2-20% (dry weight basis) depending upon when it is harvested, with an estimated average value around 10%. In addition, by providing higher quality analytical data, the new HPLC method should enable biomass researchers to obtain more accurate feedstock and process intermediate compositional data as well as better closure of carbon mass balances across pretreatment and saccharification/fermentation processes. For all of these reasons, this new HPLC method will facilitate more efficient biomass technology research, development, and deployment efforts.

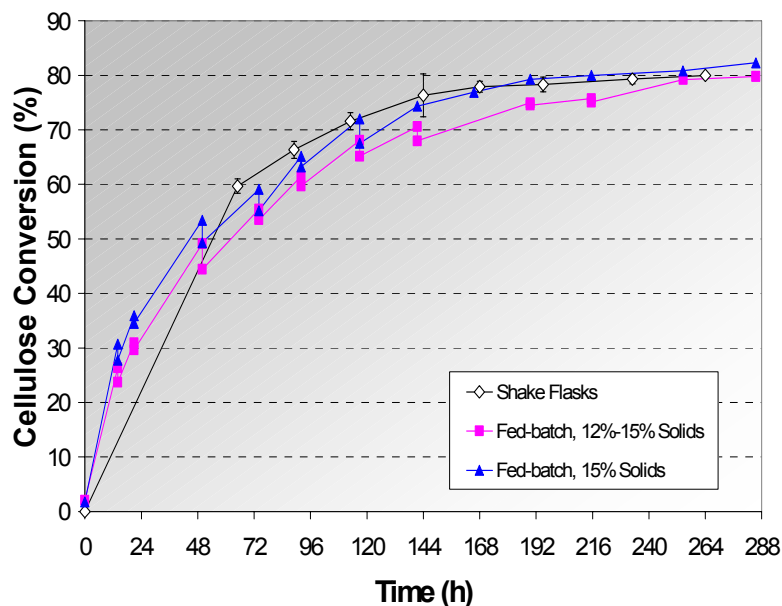
**New Steps Taken in Automating Wet Chemical Analysis of Biomass Samples.** A major bottleneck in wet chemical analysis of biomass samples is sequential solvent extraction to remove compounds that would otherwise interfere with subsequent sample analysis. To overcome this limitation, a Dionex Accelerated Solvent Extraction system was purchased and installed. By reducing sample pre-extraction times from 48 hours to 8 hours or less, this equipment will greatly accelerate wet chemical compositional analyses being performed to support a variety of Biomass Program CRADAs and core R&D projects. Ultimately, methodologies developed using this system will be incorporated into revised American Society for Testing and Materials (ASTM) standard biomass analysis methods.



## Enzymatic Cellulose Saccharification Achieved at High Solids Level in Stirred Tank Reactors.

Economic analysis has indicated that high-solids enzymatic cellulose hydrolysis will significantly reduce the operating and capital costs of ethanol production. The major process challenge posed by high solids operation is the ability to achieve high extents of conversion in a system where good heat and mass transfer are a problem. For example, a stirred tank reactor (STR) relies on good mixing to transfer heat into the bulk slurry containing solid cellulose where the reaction is performed. Previous experiments demonstrated that good

cellulose conversion could be achieved at a high insoluble solids loading (25%, w/w) using commercially available enzymes in shake flasks, where heat transfer limitations do not exist because of the uniform temperature environment. Cellulose to glucose conversion yields of 80% and 83% and glucose concentrations in the liquor phase of 125- and 140-g/L were achieved after 7 days at 20- and 40-mg/g enzyme loadings, respectively. We also recently demonstrated that performance in shake flasks and STRs is comparable at modest solids loadings (10%-15%) when the impeller configuration permits effective mixing and the heat transfer fluid temperature is maintained below 55°C. Building upon these efforts, fed-batching was used to limit the insoluble solids loading in a STR to less than 15%, while achieving a total cumulative insoluble solids loading of 25% (see figure to left). Ultimately, the level of cellulose conversion achieved in the fed-batch reactor operated in this manner was the



Cellulose conversion during enzymatic hydrolysis in batch shake flask (25% solids) and fed-batch stirred tank reactor (25% cumulative solids) at an enzyme loading of 40 mg/g. One reactor fed when insoluble solids loading decreased to 12% solids raising level to 15% (■), the other maintained at an insoluble solids loading of 15% (▲).

same as that achieved in a shake flask loaded to 25% solids. The realization of high sugar concentrations made possible by operating the key unit operations of pretreatment and saccharification at high solids loading is expected to significantly reduce the cost of biomass-derived sugars and facilitate efforts to produce realistic process residues and wastes.

## Related Activities



**New Methodology for Cellulase Enzyme Performance Testing.** A new standard procedure for assaying cellulase/cellulose conversion performance using a so-called Hybrid Hydrolysis and Fermentation (HHF) technique has recently been developed. The procedure (Laboratory Analytical Procedure) was developed to overcome temperature limitations of the current simultaneous saccharification and fermentation (SSF) approach, which is constrained by the maximum temperature that can be tolerated by the fermentative



microorganism. In the new HHF-based performance assay, the SSF stage is preceded by a “pre-saccharification” step where the fermentation microorganism is absent, allowing for greater flexibility in the range of temperatures under which enzymatic hydrolysis performance can be tested. This new assay will benefit academic and industry stakeholders by providing a method for researchers to more accurately quantify the benefits of the more thermostable, next generation cellulases now being developed.

**Process Integration Project Information.** Web-based information on the process integration project including our recent presentations at stage gate review meetings can be found at the following link ([Process Integration Project Information](#)). A discussion of how Stage Gate management is used in the Biomass Program is also available at this site ([Stage Gate Management](#)).

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